UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/518,414	08/30/2005	Johannes M. Van Den Brink	4560-4	7936

23117 7590 06/17/2009

NIXON & VANDERHYE, PC
901 NORTH GLEBE ROAD, 11TH FLOOR
ARLINGTON, VA 22203

EXAMINER	
JOIKE, MICHELE K	

ART UNIT	PAPER NUMBER
1636	

MAIL DATE	DELIVERY MODE
06/17/2009	PAPER

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

UNITED STATES PATENT AND TRADEMARK OFFICE

Commissioner for Patents
United States Patent and Trademark Office
P.O. Box 1450
Alexandria, VA 22313-1450
www.uspto.gov

BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

Application Number: 10/518,414
Filing Date: August 30, 2005
Appellant(s): VAN DEN BRINK ET AL.

Mary Wilson
For Appellant

EXAMINER'S ANSWER

This is in response to the appeal brief filed February 13, 2009 appealing from the Office action mailed May 13, 2008.

(1) Real Party In Interest

The real party of interest in this case is Chr. Hansen A/S, to whom all interest in the present application has been assigned.

(2) Related Appeals and Interferences

The examiner is not aware of any related appeals, interferences, or judicial proceedings which will directly affect or be directly affected by or have a bearing on the Board's decision in the pending appeal.

(3) Status of Claims

The statement of the status of claims contained in the brief is incorrect. A correct statement of the status of the claims is as follows:

Claims 6, 7, 20 and 21 are allowed.

Claims 1-5, 8-15, 17-19, 22 and 23 are rejected.

Claim 16 is cancelled.

(4) Status of Amendments After Final

The appellant's statement of the status of amendments after final rejection contained in the brief is correct.

(5) Summary of Claimed Subject Matter

The summary of claimed subject matter contained in the brief is correct.

(6) Grounds of Rejection to be Reviewed on Appeal

The appellant's statement of the grounds of rejection to be reviewed on appeal is correct.

(7) Claims Appendix

The copy of the appealed claims contained in the Appendix to the brief is correct.

(8) Evidence Relied Upon

6,127,142	Harboe	10-2000
5,800,849	Heldt-Hansen	9-1998

Kasturi et al "Regulation of N-linked core glycosylation: use of a site-directed mutagenesis approach to identify Asn-Xaa-Ser/Thr sequons that are poor oligosaccharide acceptors" Biochemistry, vol323 (1997), pp. 415-419

Korman et al. "Cloning, characterization, and expression of two alpha-amylase genes from Aspergillus niger var. awamori" Curr. Genet., vol17 (1990), pp. 203-212

(9) Grounds of Rejection

The following ground(s) of rejection are applicable to the appealed claims:

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

A person shall be entitled to a patent unless –

> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claims 17-19 are rejected under 35 U.S.C. 102(b) as being anticipated by US 6,127,142.

Applicants claim an active bovine chymosin comprising a N-X-T glycosylation site.

US 6,127,142 (specifically columns 1-3, and 6) teaches an aspartic protease used for clotting milk (column 1, lines 34-40). The protease should have an activity ratio similar to bovine chymosin (column 6, lines 38-41), the implication being that bovine chymosin is acceptable to use as the protease. The modified aspartic protease has a glycosylation site, which can be the sequence N-X-T (column 3, lines 38-41).

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was

not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

Claims 1-5, 8, 12-15, 17-19 and 22 are rejected under 35 U.S.C. 103(a) as being unpatentable over US 5,800,849, in view of Kasturi et al and in further view of US 6,127,142.

Applicants claim a process for producing an isolated polynucleotide sequence encoding a modified aspartic protease comprising modifying the polynucleotide sequence to encode an extra N-X-T glycosylation site in the aspartic acid protease amino acid sequence, and isolating the polynucleotide sequence. The aspartic protease is a bovine prochymosin. The modified aspartic protease can also have an artificial linker comprising the N-X-T glycosylation site. Applicants also claim the isolated polynucleotide sequence encoding the modified aspartic protease. Further, the Applicants claim a method of producing the modified aspartic protease by cultivating an *Aspergillus* cell comprising the polynucleotide sequence so that the modified aspartic protease is produced and active. Lastly, Applicants claim an active bovine chymosin comprising a N-X-T glycosylation site.

US 5,800,849 (specifically columns 1-3, Ex. 1) teaches a process for producing cheese by adding an aspartic protease to clot milk (Abstract). The process includes isolating a DNA sequence encoding a bovine prochymosin and transforming it into *Aspergillus* (column 1, lines 37-40, 54, and 55-56). The prochymosin has an N-bound glycosylation site (column 3, lines 11-15). After the DNA sequence encoding the

aspartic protease was transformed into a cell, and the protease was produced, the protease was recovered and used to make cheese, so was therefore active (Ex. 1).

US 5,800,849 does not teach the glycosylation site being N-X-T.

Kasturi et al (Biochem. J. 323: 415-419, 1997, specifically pp. 415-416) teach an N-linked glycosylation protein. The glycosylation site is N-X-T, and can be mutated by site-directed mutagenesis, and therefore be an artificial linker. However, Kasturi et al do not teach a N-linked aspartic protease.

US 6,127,142 (specifically columns 1-3, and 6) teaches an aspartic protease used for clotting milk (column 1, lines 34-40). The protease should have an activity ratio similar to bovine chymosin (column 6, lines 38-41), the implication being that bovine chymosin is acceptable to use as the protease. The modified aspartic protease has a glycosylation site, which can be the sequence N-X-T (column 3, lines 38-41).

The ordinary skilled artisan, desiring to use a N-X-T glycosylation site in chymosin would have been motivated to combine the teachings of US 5,800,849 of process of isolating a DNA sequence encoding a N-glycosylated bovine prochymosin and transforming it into *Aspergillus* with the teachings of Kasturi et al of an N-X-T linked protein and of US 6,127,142, of a modified aspartic protease that has a glycosylation site of N-X-T, because Kasturi et al teach that N-linked glycosylation usually occurs at N-X-S/T sites, and N-glycosylation profoundly affects a protein's expression and function. It would have been obvious to one of ordinary skill in the art to use a N-X-T site because N-X-T sites are generally better oligosaccharide acceptors than N-X-S sites (Kasturi et al, p. 418). Given the teachings of the prior art and the level of the

ordinary skilled artisan at the time of the applicant's invention, it must be considered, absent evidence to the contrary, that said skilled artisan would have had a reasonable expectation of success in practicing the claimed invention.

Claims 9-11 and 23 are rejected under 35 U.S.C. 103(a) as being unpatentable over US 5,800,849, Kasturi et al and US 6,127,142 as applied to claims 1-5, 8, 12-19 and 22 above, and further in view of Korman et al.

Applicants claim a process for producing an isolated polynucleotide sequence encoding a modified aspartic protease comprising modifying the polynucleotide sequence to encode an extra N-X-T glycosylation site in the aspartic acid protease amino acid sequence, and isolating the polynucleotide sequence. The aspartic protease is a bovine prochymosin. Additionally, the aspartic protease can comprise alpha-amylase as a fusion partner, wherein the artificial linker is situated between a pro-sequence and the fusion partner. Applicants also claim the isolated polynucleotide sequence encoding the modified aspartic protease. Further, the Applicants claim a method of producing the modified aspartic protease by cultivating an *Aspergillus awamori* cell comprising the polynucleotide sequence so that the modified aspartic protease is produced and active. Lastly, Applicants claim an active bovine chymosin comprising a N-X-T glycosylation site.

US 5,800849, Kasturi et al and US 6,127,142 teach all of the limitations as described above. However, they do not teach that the aspartic protease can comprise alpha-amylase as a fusion partner, wherein the artificial linker is situated between a pro-sequence and the fusion partner. They also do not teach transformation of an isolated

polynucleotide sequence encoding a modified aspartic protease in to *Aspergillus awamori.*

Korman et al (Curr. Genet. 17: 203-212, 1990, specifically pp. 203, 204, 212 and figure 5) teach a vector with a gene fusion of alpha-amylase and bovine prochymosin transformed into *Aspergillus awamori* (Materials & Methods). A synthetic DNA linker encoding the last five codons of the *amyA* gene and the first six codons of prochymosin were used to join the *amyA* and prochymosin genes (figure 5). Co-transformants were screened for chymosin production.

The ordinary skilled artisan, desiring to have an aspartic protease comprising an alpha-amylase as a fusion partner, wherein an artificial linker is situated between a pro-sequence and the fusion partner would have been motivated to combine the teachings of US 5,800,849 of process of isolating a DNA sequence encoding a N-glycosylated bovine prochymosin and transforming it into *Aspergillus*, and the teachings of Kasturi et al of an N-X-T linked protein and of US 6,127,142, of a modified aspartic protease has a glycosylation site of N-X-T, with the teachings of Korman et al teaching a gene fusion of alpha-amylase and bovine prochymosin with a synthetic DNA linker encoding the last five codons of the *amyA* gene and the first six codons of prochymosin because Korman et al teach that the alpha-amylase gene promoter has been used to derive high level expression of an aspartic protease gene from fungi (p. 203). It would have been obvious to one of ordinary skill in the art to use alpha-amylase as a fusion partner because gene fusions are useful in studying the regulation of genes, and the potential use of *amyA* transcriptional and translational control elements in combination with

aspartic proteases has been demonstrated (p. 212). Given the teachings of the prior art and the level of the ordinary skilled artisan at the time of the applicant's invention, it must be considered, absent evidence to the contrary, that said skilled artisan would have had a reasonable expectation of success in practicing the claimed invention.

(10) Response to Argument

US 6,127,142 (specifically columns 1-3, and 6) teaches an aspartic protease used for clotting milk (column 1, lines 34-40). The protease should have an activity ratio similar to bovine chymosin (column 6, lines 38-41), the implication being that bovine chymosin is acceptable to use as the protease, since nothing is closer to bovine chymosin, than bovine chymosin. Appellants argue that the reference teaches an aspartic protease that is not chymosin. However, they do not exclude chymosin. Appellants are citing EC 3.4.23.23 as the aspartic protease taught in US 6,127,142. This aspartic protease is taught in Ex. 1, which also teaches the enzyme, Hannilase™, which is a member of the chymosin family. US 6,127,142 anticpates claims 17-19.

In response to appellants' argument that there is no suggestion to combine the references, the examiner recognizes that obviousness can only be established by combining or modifying the teachings of the prior art to produce the claimed invention where there is some teaching, suggestion, or motivation to do so found either in the references themselves or in the knowledge generally available to one of ordinary skill in the art. See *In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988)and *In re Jones*, 958 F.2d 347, 21 USPQ2d 1941 (Fed. Cir. 1992). In this case, US 6,127,142

teaches the use of an extra NXT glycosylation site. Kasturi et al teach that the glycosylation site is N-X-T, and can be mutated by site-directed mutagenesis. Together with US 5,800,849, they teach modifying the polynucleotide sequence to encode an extra NXT glycosylation site (see previous office action).

Appellants are arguing that US 5,800,849 teach increased production of cheese is due to increased glycosylation, Kasturi et al teach the impact of NXT sites, and US 6,127,142 teaches that improved milk clotting activity is due to decreased glycosylation. the references are inconsistent and not combinable.

However, US 6,127,142 teaches the use of chymosin, and an NXT site present on a protease. While it does teach deglycosylating proteases, it does not teach complete deglycosylation. "The above treatment according to the invention results in removal of glycosyl moieties to an extent which depends on the degree of glycosylation and, when a deglycosylating enzyme is applied, the amount and the enzymatic activity of the enzyme and on the time of enzyme treatment. As an example, the coagulant produced natively by Rhizomucor miehei has three possible N-linked glycosylation sites i.e. sites having the sequence Asn-X-Thr/Ser at Asn.sup.79, Asn.sup.188 and Asn.sup.313, but it has been found that only two of these sites (79, 188) are glycosylated (Boel et al., 1986, Genetics, 1, 363-369). However, even if two possible glycosylation sites can be glycosylated by the homologous strain or a heterologous strain, the degree with which these sites are glycosylated may vary, e.g. according to the producing strain and the growth medium and conditions.'" col3., lines 34-48. "In accordance with the invention it is preferred that essentially all of the glycosyl moieties

initially present in Rhizomucor miehei aspartic protease are removed, although it is expected that an enhancement of the milk clotting activity of such an enzyme will be found at lower degrees of glycosyl removal. Thus, the method according to the invention will preferably result in at least 10% deglycosylation, such as at least 20%. In more preferred embodiments, at least 50% of the glycosyl groups are removed such as least 75%." col. 3, lines 53-62. In other words, complete deglycosylation does not occur, and some glycosyl groups will remain and affect activity. While fewer glycosyl groups may cause an increase in activity, there are still glycosyl (NXT) groups present. So, the enzyme is not completely deglycosylated to increase activity. Furthermore, Kasturi et al teach that N-linked glycosylation usually occurs at N-X-S/T sites, and N-glycosylation profoundly affects a protein's expression and function, and US 5,800,849, col1, lines 25-27, teach an increase in cheese yield by glycosylating the protease, as noted by Appellants.

Appellants argue that the present invention results from Appellants' desire to obtain the enzyme in higher yield. First of all, nothing in the claims indicate that a higher yield is achieved or desired. Secondly, the Examiner has a different motivation or "purpose" for combining the references as noted in the September 18, 2007 office action. The fact that the Examiner does not have the same reason as the Applicants for creating the invention, does not make the Examiner's reasoning hindsight.

(11) Related Proceeding(s) Appendix

No decision rendered by a court or the Board is identified by the examiner in the Related Appeals and Interferences section of this examiner's answer.

For the above reasons, it is believed that the rejections should be sustained.

Respectfully submitted,

Michele K. Joike

Conferees:

/NANCY VOGEL/

Primary Examiner, Art Unit 1636

/ Christopher S. F. Low /
Supervisory Patent Examiner, Art Unit 1636

/Nashaat T. Nashed/
Primary Examiner, Art Unit 1656